

FIGURE 1

Relative TS gene expression in matching primary and metastatic tissue in CRC (both paraffin)

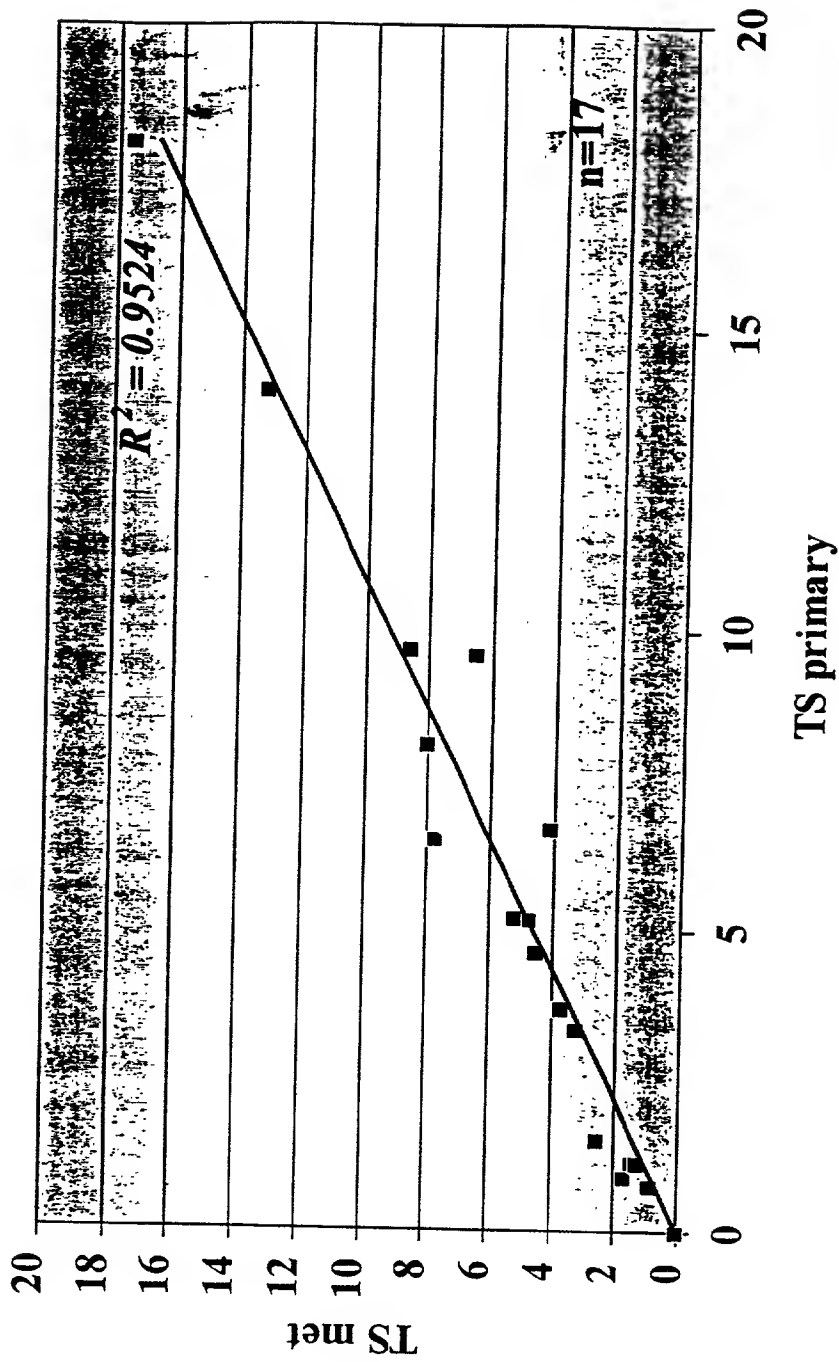


Figure 2: Chart illustrating how to calculate *EGFR* expression relative to an internal control gene

Sample	from "test" reactions				from "calibration" reactions				Uncorrected Gene Expression (UGE)	Known <i>EGFR</i> values	Derivation of <i>K_{EGFR}</i> (average <i>K</i>)	Relative <i>EGFR</i> exp
	<i>C_T</i> <i>EGFR</i>	<i>C_T</i> <i>β-actin</i>	ΔC_T	$2^{-\Delta C_T}$	<i>C_T</i> <i>EGFR</i>	<i>C_T</i> <i>β-actin</i>	ΔC_T	$2^{-\Delta C_T}$				
Experimental	unknown 1	32.7	26.8	5.9	0.0167	-	-	-	0.525	-	26.95×10^3	14.4×10^3
	unknown 2	32.88	26.43	6.45	0.0114	-	-	-	0.358	-	26.95×10^3	9.66×10^3
	Calib. RNA	-	-	-	-	22.04	4.97	0.0319	$0.0319 \times 0.0319 = 1$	-	-	-
From Published Data	60N	31.61	23.86	7.75	0.00464	-	-	-	0.2117	5.70×10^3	26.95×10^3	-
	60T	29.08	20.65	8.43	0.0029	-	-	-	0.1321	3.56×10^3	26.95×10^3	-
	SF12A	28.71	20.76	7.95	0.0040	-	-	-	0.184	4.97×10^3	26.95×10^3	-
	SF12B	24.69	19.87	4.82	0.0354	-	-	-	1.613	43.5×10^3	26.95×10^3	-
	Cir11	24.03	16.3	7.73	0.0047	-	-	-	0.215	5.78×10^3	26.95×10^3	-
	AdCol	26.04	17.06	8.98	0.00198	-	-	-	0.090	2.43×10^3	26.95×10^3	-
	Calib. RNA	-	-	-	-	26.96	7.39	0.00596	$0.00596 / 0.00596 = 1$	-	-	-

Fig. 3. Chart illustrating how to calculate *DPD* expression relative to an internal control gene.

	From Test Reaction				From Calibrator Reaction				Undiluted Calibrator Expression (log ₂ F)	Published <i>DPD</i> Value	Log ₂ Ratio of <i>DPD</i> to K	Corrected Relative <i>DPD</i> Exp.
	Sample	<i>C_T</i> <i>DPD</i>	<i>C_T</i> β-actin	ΔC_T	$2^{-\Delta C_T}$	<i>C_T</i> calib. RNA	<i>C_T</i> β-actin	ΔC_T	$2^{-\Delta C_T}$			
Experimental	unknown 1	25.05	19.84	5.21	.027	-	-	-	4.47	-	1.08×10^{-3}	4.83×10^{-3}
	unknown 2	28.18	18.76	9.42	.0015	-	-	-	0.241	-	1.08×10^{-3}	0.2608×10^{-3}
	Calib. RNA	-	-	-	-	26.92	19.55	7.37	.006	0.006/0.006=1	-	1.08×10^{-3}
From Published Data	A	31.04	24.56	6.49	.0111	-	-	-	2.45	2.7×10^{-3}	1.10×10^{-3}	-
	B	27.95	20.5	7.45	.0057	-	-	-	1.26	1.2×10^{-3}	1.08×10^{-3}	-
	C	26.88	19.2	7.68	.00488	-	-	-	1.07	1.1×10^{-3}	1.02×10^{-3}	-
	D	33.32	22.88	10.44	.00072	-	-	-	0.158	0.17×10^{-3}	1.08×10^{-3}	-
	E	26.96	22.01	4.95	.03235	-	-	-	7.12	7.3×10^{-3}	1.03×10^{-3}	-
	F	25.44	21.4	4.04	.0607	-	-	-	13.38	16×10^{-3}	1.2×10^{-3}	-
	Calib. RNA	-	-	-	-	27.88	20.098	7.782	0.005	0.005/0.005 = 1	-	-

Figure 4: Chart illustrating how to calculate *TS* expression relative to an internal control gene

	From raw reaction					From calibration (2507)					Uncertainty Estimation $2 \times 10^{-2} \Delta$ 0.055		Published data		Derivation of K_{TS} (average)		Relative TS_{90}	
	Sample	C_T TS	C_T house	ΔC_T	$2^{-\Delta C_T}$	C_T TS	C_T house	ΔC_T	$2^{-\Delta C_T}$	$2^{-\Delta C_T}$	$2 \times 10^{-2} \Delta$ 0.055	$2 \times 10^{-2} \Delta$ 0.055	K_{TS}	K_{TS}	K_{TS}	K_{TS}	K_{TS}	K_{TS}
Experimental	unknown 1	26.14	19.35	6.79	0.00903	-	-	-	-	-	0.178	-	-	-	12.8×10^3	12.8×10^3	2.25×10^3	-
	unknown 2	32.07	28.38	3.69	0.0748	-	-	-	-	-	1.33	-	-	-	12.6×10^3	12.6×10^3	16.758×10^3	-
	Calib. RNA	-	-	-	-	27.94	23.79	4.15	0.0563	0.056/0.056=1	-	-	-	-	-	-	-	-
From Published Data	L7	26.94	24.55	2.39	0.191	-	-	-	-	-	3.18	-	38.8×10^3	12.2×10^3	12.6×10^3	12.6×10^3	-	-
	L91	24.91	22.12	2.79	0.144	-	-	-	-	-	2.40	-	29.55×10^3	12.31×10^3	12.6×10^3	12.6×10^3	-	-
	L121	24.95	20.89	4.06	0.059	-	-	-	-	-	0.88	-	12.22×10^3	13.88×10^3	12.6×10^3	12.6×10^3	-	-
	L150	26.77	22.88	6.89	0.008	-	-	-	-	-	0.133	-	1.72×10^3	12.93×10^3	12.6×10^3	12.6×10^3	-	-
	L220	26.52	19.77	6.75	0.0092	-	-	-	-	-	0.153	-	1.89×10^3	12.35×10^3	12.6×10^3	12.6×10^3	-	-
	L164	26.81	21.21	5.6	0.0205	-	-	-	-	-	0.341	-	4.2×10^3	12.31×10^3	12.6×10^3	12.6×10^3	-	-
	Calib. RNA	-	-	-	-	25.14	20.09	5.04	0.06	0.06/0.06=1	-	-	-	-	-	-	-	-